

WHAT WE CLAIM:

1. A method for detecting a target nucleic acid sequence in a sample, the sample comprising nucleic acid molecules of higher biological complexity relative to amplified nucleic acid molecules and the target nucleic acid sequence differs from one or more nucleic acid sequences by at least one nucleotide, the method comprising the steps of:
 - a) providing an addressable substrate having a capture oligonucleotide bound thereto, wherein the capture oligonucleotide has a sequence that is complementary to at least part of a first portion of the target nucleic acid sequence;
 - b) providing a detection probe comprising detector oligonucleotides, wherein the detector oligonucleotides have a sequence that is complementary to at least part of a second portion of the target nucleic acid sequence of step (a);
 - c) contacting the sample with the substrate and the detection probe under conditions that are effective for the hybridization of the capture oligonucleotide to the first portion of the target nucleic acid sequence and the hybridization of the detection probe to the second portion of the target nucleic acid sequence and to allow for discrimination between the target and said one or more nucleic acid sequences that differ by at least one nucleotide; and
 - d) detecting whether the capture oligonucleotide and detection probe hybridized with the first and second portions of the target nucleic acid sequence.

2. The method of claim 1, wherein the target nucleic acid sequence comprises a Single Nucleotide Polymorphism.

3. The method of claim 1, wherein the single nucleotide difference is recognized by the capture oligonucleotide bound to the substrate.

4. The method of claim 1, wherein the single nucleotide difference is recognized by the detector oligonucleotides.

5. The method of claim 1, wherein the target nucleic acid molecules comprise genomic DNA, genomic RNA, expressed RNA, plasmid DNA, mitochondrial or other cell organelle DNA, free cellular DNA, viral DNA or viral RNA, or a mixture of two or more of the above.

6. The method of claim 1, wherein the substrate comprises a plurality of capture oligonucleotides, each of which can recognize a different single nucleotide polymorphism.

7. The method of claim 1, wherein the sample comprises more than one nucleic acid target, each of which comprises one or more different single nucleotide polymorphisms.

8. The method of claim 1, wherein one or more types of detector probes are provided, each of which has detector oligonucleotides bound thereto that are capable of hybridizing with a different nucleic acid target.

9. The method of claim 1, wherein sample is contacted with the detector probe so that a nucleic acid target present in the sample hybridizes with the detector oligonucleotides on the detector probe, and the nucleic acid target bound to the detector probe is then contacted with the substrate so that the nucleic acid target hybridizes with the capture oligonucleotide on the substrate.

10. The method of claim 1, wherein sample is contacted with the substrate so that a nucleic acid target present in the sample hybridizes with a capture oligonucleotide, and the nucleic acid target bound to the capture oligonucleotide is then contacted with the detector probe so that the nucleic acid target hybridizes with the detector oligonucleotides on the detector probe.

11. The method of claim 1, wherein the sample is contacted simultaneously with the detector probe and the substrate.

12. The method of claim 1, wherein the detector oligonucleotides comprise a detectable label.

13. The method of claim 12, wherein the detection label allows detection by photonic, electronic, acoustic, opto-acoustic, gravity, electro-chemical, electro-optic, mass-spectrometric, enzymatic, chemical, biochemical, or physical means.

14. The method of claim 12, wherein the label is fluorescent.

25

15. The method of claim 12, wherein the label is luminescent.
16. The method of claim 12, wherein the label is phosphorescent.
- 5 17. The method of claim 12, wherein the label is radioactive.
18. The method of claim 12, wherein the label is a nanoparticle.
19. The method of claim 12, wherein the label is a dendrimer.
- 10 20. The method of claim 12, wherein the label is a molecular aggregate.
21. The method of claim 12, wherein the label is a quantum dot.
- 15 22. The method of claim 12, wherein the label is a bead.
23. The method of claim 1, wherein the detector probe is a nanoparticle probe having detector oligonucleotides bound thereto.
- 20 24. The method of claim 23, wherein the nanoparticles are made of a noble metal.
25. The method of claim 24, wherein the nanoparticles are made of gold or silver.

25

26. The method of claim 25, wherein the nanoparticles are made of gold.

27. The method of claim 23, wherein the detecting comprises contacting the substrate with silver stain.

5 28. The method of claim 23, wherein the detecting comprises detecting light scattered by the nanoparticle.

29. The method of claim 23, wherein the detecting comprises observation with an optical scanner.

10

30. The method of claim 23, wherein the detecting comprises observation with a flatbed scanner.

31. The method of claim 29 or 30, wherein the scanner is linked to a computer
15 loaded with software capable of calculating grayscale measurements, and the grayscale measurements are calculated to provide a quantitative measure of the amount of nucleic acid detected.

32. The method of claim 23, wherein the oligonucleotides attached to the
20 substrate are located between two electrodes, the nanoparticles are made of a material that is a conductor of electricity, and step (d) comprises detecting a change in conductivity.

33. The method of claim 32, wherein the electrodes are made of gold and the nanoparticles are made of gold.

25

34. The method of claim 32, wherein the substrate is contacted with silver stain to produce the change in conductivity.

35. The method of claims 23, wherein a plurality of oligonucleotides, each of which can recognize a different target nucleic acid sequence, are attached to the substrate in an array of spots and each spot of oligonucleotides is located between two electrodes, the nanoparticles are made of a material that is a conductor of electricity, and step (d) comprises detecting a change in conductivity.

36. The method of claim 35, wherein the electrodes are made of gold and the nanoparticles are made of gold.

37. The method of claim 35, wherein the substrate is contacted with silver stain to produce the change in conductivity.

15

38. A method for detecting one or more target nucleic acid sequences in a sample, the sample comprising nucleic acid molecules of higher biological complexity relative to amplified nucleic acid molecules and the one or more target nucleic acid sequences each differ from known nucleic acid sequences by at least one nucleotide, the method comprising the steps of:

- a) providing an addressable substrate having a plurality of capture oligonucleotides bound thereto, wherein the capture oligonucleotides have sequences that are complementary to one or more portions of the one or more target nucleic acid sequences;

- b) providing one or more detector probes comprising detector oligonucleotides, wherein the detector oligonucleotides have sequences that are complementary to one or more portions of the one or more target nucleic acid sequences of step (a) that are not recognized by a capture oligonucleotide on the substrate;
- c) contacting the sample with the substrate and the detector probes under conditions that are effective for the hybridization of the capture oligonucleotides to one or more portions of the one or more target nucleic acid sequences and the hybridization of the detector probes to portions of the one or more target nucleic acid sequences that are not recognized by a capture oligonucleotide and to allow for discrimination between targets that differ by at least one nucleotide; and
- d) detecting whether any of the capture oligonucleotide and detector probes hybridized with any of the target nucleic acid sequences.

39. The method of claim 38, wherein the target nucleic acid sequence comprises a Single Nucleotide Polymorphism.

40. The method of claim 38, wherein the single nucleotide difference is recognized by the capture oligonucleotide bound to the substrate.

41. The method of claim 38, wherein the single nucleotide difference is recognized by the detector oligonucleotides.

42. The method of claim 38, wherein the target nucleic acid molecules comprise genomic DNA, genomic RNA, expressed RNA, plasmid DNA, mitochondrial or other cell organelle DNA, free cellular DNA, viral DNA or viral RNA, or a mixture of two or more of the above.

5

43. The method of claim 38, wherein the substrate comprises a plurality of capture oligonucleotides, each of which can recognize a different single nucleotide polymorphism.

10

44. The method of claim 38, wherein the sample comprises more than one nucleic acid target, each of which comprises a different single nucleotide polymorphism.

15

45. The method of claim 38, wherein one or more types of detector probes are provided, each of which has detector oligonucleotides bound thereto that are capable of hybridizing with a different nucleic acid target.

20

46. The method of claim 38, wherein sample is contacted with the detector probe so that a nucleic acid target present in the sample hybridizes with the detector oligonucleotides on the detector probe, and the nucleic acid target bound to the detector probe is then contacted with the substrate so that the nucleic acid target hybridizes with the capture oligonucleotide on the substrate.

25

47. The method of claim 38, wherein sample is contacted with the substrate so that a nucleic acid target present in the sample hybridizes with a capture oligonucleotide, and the nucleic acid target bound to the capture oligonucleotide is then contacted with the

detector probe so that the nucleic acid target hybridizes with the detector oligonucleotides on the detector probe.

48. The method of claim 38, wherein the sample is contacted simultaneously
5 with the detector probe and the substrate.

49. The method of claim 38, wherein the detector probe comprise a detectable label.

10 50. The method of claim 49, wherein the detection label allows detection by photonic, electronic, acoustic, opto-acoustic, gravity, electro-chemical, electro-optic, mass-spectrometric, enzymatic, chemical, biochemical, or physical means.

15 51. The method of claim 49, wherein the label is fluorescent.

52. The method of claim 49, wherein the label is luminescent.

53. The method of claim 49, wherein the label is phosphorescent.

20 54. The method of claim 49, wherein the label is radioactive.

55. The method of claim 49, wherein the label is a nanoparticle.

25 56. The method of claim 49, wherein the label is a dendrimer.

57. The method of claim 49, wherein the label is a molecular aggregate.
58. The method of claim 49, wherein the label is a quantum dot.
- 5 59. The method of claim 49, wherein the label is a bead.
60. The method of claim 38, wherein the detector probe is a nanoparticle probe having detector oligonucleotides bound thereto.
- 10 61. The method of claim 60, wherein the nanoparticles are made of a noble metal.
62. The method of claim 61, wherein the nanoparticles are made of gold or silver.
- 15 63. The method of claim 62, wherein the nanoparticles are made of gold.
64. The method of claim 60, wherein the detecting comprises contacting the substrate with silver stain.
- 20 65. The method of claim 60, wherein the detecting comprises observation of light scattered by the nanoparticle.
66. The method of claim 60, wherein the detecting comprises observation with an optical scanner.

67. The method of claim 60, wherein the detecting comprises observation with a flatbed scanner.

68. The method of claim 66 or 67, wherein the scanner is linked to a computer
5 loaded with software capable of calculating grayscale measurements, and the grayscale measurements are calculated to provide a quantitative measure of the amount of nucleic acid detected.

69. The method of claim 60, wherein the oligonucleotides attached to the
10 substrate are located between two electrodes, the nanoparticles are made of a material that is a conductor of electricity, and step (d) comprises detecting a change in conductivity.

70. The method of claim 69, wherein the electrodes are made of gold and the nanoparticles are made of gold.
15

71. The method of claim 69, wherein the substrate is contacted with silver stain to produce the change in conductivity.

72. The method of claims 60, wherein a plurality of oligonucleotides, each of
20 which can recognize a different target nucleic acid sequence, are attached to the substrate in an array of spots and each spot of oligonucleotides is located between two electrodes, the nanoparticles are made of a material that is a conductor of electricity, and step (d) comprises detecting a change in conductivity.

73. The method of claim 72, wherein the electrodes are made of gold and the nanoparticles are made of gold.

74. The method of claim 72, wherein the substrate is contacted with silver stain to produce the change in conductivity.

75. A method for identifying a single nucleotide polymorphism in a sample, the sample comprising nucleic acid molecules of higher biological complexity relative to amplified nucleic acid molecules, the method comprising the steps of:

- 10 a) providing an addressable substrate having at least one capture oligonucleotide bound thereto, wherein the at least one capture oligonucleotide has a sequence that is complementary to at least a part of a nucleic acid target that comprises a specific polymorphism;
- 15 b) providing a detector probe having detector oligonucleotides bound thereto, wherein the detector oligonucleotides has a sequence that is complementary to at least a portion of the nucleic acid target of step (a);
- c) contacting the sample with the substrate and the detector probe under conditions that are effective for the hybridization of the capture oligonucleotide to the nucleic acid target and the hybridization of the detector probe to the nucleic acid target and to allow for discrimination between targets that differ by a single nucleotide; and
- 20 d) detecting whether the capture oligonucleotide and detector probe hybridized with the nucleic acid target.

76. The method of claim 75, wherein the polymorphism is recognized by the capture oligonucleotide bound to the substrate.

77. The method of claim 75, wherein the polymorphism is recognized by the
5 detector oligonucleotides.

78. The method of claim 75, wherein the nucleic acid molecules in the sample comprise genomic DNA, genomic RNA, expressed RNA, plasmid DNA, mitochondrial or other cell organelle DNA, free cellular DNA, viral DNA or viral RNA, or a mixture of
10 two or more of the above.

79. The method of claim 75, wherein the substrate comprises a plurality of capture oligonucleotides, each of which can recognize a different single nucleotide polymorphism.
15

80. The method of claim 75, wherein the sample comprises more than one nucleic acid targets, each of which comprises one or more different single nucleotide polymorphisms.

20 81. The method of claim 75, wherein one or more types of detector probes are provided, each of which has detector oligonucleotides bound thereto that are capable of hybridizing with a different nucleic acid target.

82. The method of claim 75, wherein sample is contacted with the detector
25 probe so that a nucleic acid target present in the sample hybridizes with the detector

oligonucleotides on the detector probe, and the nucleic acid target bound to the detector probe is then contacted with the substrate so that the nucleic acid target hybridizes with the capture oligonucleotide on the substrate.

5 83. The method of claim 75, wherein sample is contacted with the substrate so that a nucleic acid target present in the sample hybridizes with a capture oligonucleotide, and the nucleic acid target bound to the capture oligonucleotide is then contacted with the detector probe so that the nucleic acid target hybridizes with the detector oligonucleotides on the detector probe.

10

84. The method of claim 75, wherein the sample is contacted simultaneously with the detector probe and the substrate.

15 85. The method of claim 75, wherein the detector probes comprise a detectable label.

20 86. The method of claim 85, wherein the detection label allows detection by photonic, electronic, acoustic, opto-acoustic, gravity, electro-chemical electro-optic, mass-spectrometric, enzymatic, chemical, biochemical, or physical means.

25

87. The method of claim 85, wherein the label is fluorescent.

88. The method of claim 85, wherein the label is a nanoparticle.

25 89. The method of claim 85, wherein the label is luminescent.

- 5
90. The method of claim 85, wherein the label is phosphorescent.
91. The method of claim 85, wherein the label is radioactive.
92. The method of claim 85, wherein the label is a dendrimer.
93. The method of claim 85, wherein the label is a molecular aggregate.
- 10 94. The method of claim 85, wherein the label is a quantum dot.
95. The method of claim 85, wherein the label is a bead.
- 15 96. The method of claim 75, wherein the detector probe is a nanoparticle probe having detector oligonucleotides bound thereto.
97. The method of claim 96, wherein the nanoparticles are made of a noble metal.
- 20 98. The method of claim 97, wherein the nanoparticles are made of gold or silver.
99. The method of claim 98, wherein the nanoparticles are made of gold.

100. The method of claim 96, wherein the detecting comprises contacting the substrate with silver stain.

101. The method of claim 96, wherein the detecting comprises detecting light
5 scattered by the nanoparticle.

102. The method of claim 96, wherein the detecting comprises observation with an optical scanner.

10 103. The method of claim 96, wherein the detecting comprises observation with a flatbed scanner.

104. The method of claim 102 or 103, wherein the scanner is linked to a computer loaded with software capable of calculating grayscale measurements, and the
15 grayscale measurements are calculated to provide a quantitative measure of the amount of nucleic acid detected.

105. The method of claim 96, wherein the oligonucleotides attached to the substrate are located between two electrodes, the nanoparticles are made of a material that
20 is a conductor of electricity, and step (d) comprises detecting a change in conductivity.

106. The method of claim 105, wherein the electrodes are made of gold and the nanoparticles are made of gold.

107. The method of claim 105, wherein the substrate is contacted with silver stain to produce the change in conductivity.

108. The method of claims 96, wherein a plurality of oligonucleotides, each of which can recognize a different single nucleotide polymorphism, are attached to the substrate in an array of spots and each spot of oligonucleotides is located between two electrodes, the nanoparticles are made of a material that is a conductor of electricity, and step (d) comprises detecting a change in conductivity.

109. The method of claim 108, wherein the electrodes are made of gold and the nanoparticles are made of gold.

110. The method of claim 109, wherein the substrate is contacted with silver stain to produce the change in conductivity.

111. A method for identifying one or more single nucleotide polymorphisms in a sample, the sample comprising nucleic acid molecules of higher biological complexity relative to amplified nucleic acid molecules, the method comprising the steps of:

- a) providing an addressable substrate having a plurality of capture oligonucleotides bound thereto, wherein the capture oligonucleotides have sequences that are complementary to multiple portions of a nucleic acid target, each said portion comprising a specific polymorphism;
- b) providing one or more detector probes comprising detector oligonucleotides, wherein the detector oligonucleotides have a sequence that is complementary to at least a portion of one of the nucleic acid targets

of step (a) that is not recognized by a capture oligonucleotide on the substrate;

- c) contacting the sample with the substrate and the detector probes under conditions that are effective for the hybridization of the capture oligonucleotides to multiple portions of the nucleic acid target and the hybridization of the detector probe to the nucleic acid target and to allow for discrimination between targets that differ by a single nucleotide; and
- d) detecting whether any of the capture oligonucleotides and detector probes hybridized with any of the nucleic acid targets.

112. The method of claim 111, wherein the polymorphism is recognized by the capture oligonucleotide bound to the substrate.

113. The method of claim 111, wherein the polymorphism is recognized by the detector oligonucleotides.

114. The method of claim 111, wherein the nucleic acid molecules in the sample comprise genomic DNA, genomic RNA, expressed RNA, plasmid DNA, mitochondrial or other cell organelle DNA, free cellular DNA, viral DNA or viral RNA, or a mixture of two or more of the above.

115. The method of claim 111, wherein the substrate comprises a plurality of capture oligonucleotides, each of which can recognize one or more different single nucleotide polymorphisms.

116. The method of claim 111, wherein the sample comprises more than one nucleic acid targets, each of which comprises a different single nucleotide polymorphism.

117. The method of claim 111, wherein one or more types of detector probes
5 are provided, each of which has detector oligonucleotides bound thereto that are capable of hybridizing with a different nucleic acid target.

118. The method of claim 111, wherein sample is contacted with the detector probe so that a nucleic acid target present in the sample hybridizes with the detector
10 oligonucleotides on the detector probe, and the nucleic acid target bound to the detector probe is then contacted with the substrate so that the nucleic acid target hybridizes with the capture oligonucleotide on the substrate.

119. The method of claim 111, wherein sample is contacted with the substrate
15 so that a nucleic acid target present in the sample hybridizes with a capture oligonucleotide, and the nucleic acid target bound to the capture oligonucleotide is then contacted with the detector probe so that the nucleic acid target hybridizes with the detector oligonucleotides on the detector probe.

20 120. The method of claim 111, wherein the sample is contacted simultaneously with the detector probe and the substrate.

121. The method of claim 111, wherein the detector oligonucleotides comprise a detectable label.

25

122. The method of claim 121, wherein the detection label allows detection by photonic, electronic, acoustic, opto-acoustic, gravity, electro-chemical, electro-optic, mass-spectrometric, enzymatic, chemical, biochemical, or physical means.

5 123. The method of claim 121, wherein the label is fluorescent.

124. The method of claim 121, wherein the label is luminescent.

125. The method of claim 121, wherein the label is phosphorescent.

10

126. The method of claim 121, wherein the label is radioactive.

127. The method of claim 121, wherein the label is a nanoparticle.

15

128. The method of claim 121, wherein the label is a dendrimer.

129. The method of claim 121, wherein the label is a molecular aggregate.

130. The method of claim 121, wherein the label is a quantum dot.

20

131. The method of claim 121, wherein the label is a bead.

132. The method of claim 111, wherein the detector probe is a nanoparticle probe having detector oligonucleotides bound thereto.

25

133. The method of claim 132, wherein the nanoparticles are made of a noble metal.

134. The method of claim 133, wherein the nanoparticles are made of gold or silver.

135. The method of claim 134, wherein the nanoparticles are made of gold.

136. The method of claim 132, wherein the detecting comprises contacting the substrate with silver stain.

137. The method of claim 132, wherein the detecting comprises detecting light scattered by the nanoparticle.

138. The method of claim 132, wherein the detecting comprises observation with an optical scanner.

139. The method of claim 132, wherein the detecting comprises observation with a flatbed scanner.

140. The method of claim 138 or 139, wherein the scanner is linked to a computer loaded with software capable of calculating grayscale measurements, and the grayscale measurements are calculated to provide a quantitative measure of the amount of nucleic acid detected.

141. The method of claim 132, wherein the oligonucleotides attached to the substrate are located between two electrodes, the nanoparticles are made of a material that is a conductor of electricity, and step (d) comprises detecting a change in conductivity.

5 142. The method of claim 141, wherein the electrodes are made of gold and the nanoparticles are made of gold.

143. The method of claim 141, wherein the substrate is contacted with silver stain to produce the change in conductivity.

10

144. The method of claims 141, wherein a plurality of oligonucleotides, each of which can recognize a different single nucleotide polymorphism, are attached to the substrate in an array of spots and each spot of oligonucleotides is located between two electrodes, the nanoparticles are made of a material that is a conductor of electricity, and
15 step (d) comprises detecting a change in conductivity.

145. The method of claim 144, wherein the electrodes are made of gold and the nanoparticles are made of gold.

20 146. The method of claim 146, wherein the substrate is contacted with silver stain to produce the change in conductivity.

147. The method of claim 1, 38, 75 or 111, wherein the higher biological complexity is greater than about 50,000.

25

148. The method of claim 1, 38, 75 or 111, wherein the higher biological complexity is between about 50,000 and about 50,000,000,000.

149. The method of claim 1, 38, 75 or 111, wherein the higher biological
5 complexity is about 1,000,000,000.

150. The method of claim 1 or 38, wherein the target nucleic acid sequence is a portion of a gene of a biological organism.

10 151. The method of claim 1 or 38, wherein the target nucleic acid sequence is a portion of a gene of a *Staphylococcus* bacterium.

152. The method of claim 151, wherein the *Staphylococcus* bacterium is *S. aureus*, *S. haemolyticus*, *S. epidermidis*, *S. lugdunensis*, *S. hominis*, or *S. saprophyticus*.

15 153. The method of claim 151, wherein the target nucleic acid sequence is a portion of the *Tuf* gene.

154. The method of claim 151, wherein the target nucleic acid sequence is a portion of the *femA* gene.

20

155. The method of claim 151, wherein the target nucleic acid sequence is a portion of the 16S rRNA gene.

156. The method of claim 151, wherein the target nucleic acid sequence is a
25 portion of the *hsp60* gene.

157. The method of claim 151, wherein the target nucleic acid sequence is a portion of the *sodA* gene.

5 158. The method of claim 1 or 38, wherein the target nucleic acid sequence is a portion of the *mecA* gene.

159. The method of claim 1 or 38, wherein the target nucleic acid sequence comprises the sequence set forth in SEQ ID NO: 17, SEQ ID NO: 18, SEQ ID NO: 19,
10 SEQ ID NO: 20, SEQ ID NO: 21, SEQ ID NO: 22, SEQ ID NO: 23, SEQ ID NO: 24, SEQ ID NO: 24, SEQ ID NO: 26, SEQ ID NO: 27, SEQ ID NO: 28, SEQ ID NO: 29, SEQ ID NO: 30, SEQ ID NO: 31, SEQ ID NO: 32, SEQ ID NO: 33, SEQ ID NO: 34, SEQ ID NO: 35, SEQ ID NO: 36, SEQ ID NO: 37, SEQ ID NO: 38, SEQ ID NO: 39, SEQ ID NO: 40, SEQ ID NO: 41, SEQ ID NO: 42, SEQ ID NO: 43, SEQ ID NO: 44,
15 SEQ ID NO: 45, SEQ ID NO: 46, SEQ ID NO: 47, SEQ ID NO: 48, SEQ ID NO: 49, SEQ ID NO: 50, SEQ ID NO: 51, SEQ ID NO: 52, SEQ ID NO: 53, SEQ ID NO: 54, SEQ ID NO: 55, SEQ ID NO: 56, SEQ ID NO: 57, SEQ ID NO: 58, SEQ ID NO: 59, SEQ ID NO: 60, SEQ ID NO: 61, SEQ ID NO: 62, SEQ ID NO: 63, SEQ ID NO: 64, SEQ ID NO: 65, SEQ ID NO: 66, SEQ ID NO: 67, SEQ ID NO: 68, SEQ ID NO: 69,
20 SEQ ID NO: 70, SEQ ID NO: 71, SEQ ID NO: 72, SEQ ID NO: 73, SEQ ID NO: 74, SEQ ID NO: 75, SEQ ID NO: 76, SEQ ID NO: 77, or SEQ ID NO: 78.

160. The method of claim 1 or 38, wherein at least one of the detection oligonucleotides comprise the sequence set forth in SEQ ID NO: 17, SEQ ID NO: 18,
25 SEQ ID NO: 19, SEQ ID NO: 20, SEQ ID NO: 21, SEQ ID NO: 22, SEQ ID NO: 23,

SEQ ID NO: 24, SEQ ID NO: 24, SEQ ID NO: 26, SEQ ID NO: 27, SEQ ID NO: 28,
SEQ ID NO: 29, SEQ ID NO: 30, SEQ ID NO: 31, SEQ ID NO: 32, SEQ ID NO: 33,
SEQ ID NO: 34, SEQ ID NO: 35, SEQ ID NO: 36, SEQ ID NO: 37, SEQ ID NO: 38,
SEQ ID NO: 39, SEQ ID NO: 40, SEQ ID NO: 41, SEQ ID NO: 42, SEQ ID NO: 43,
5 SEQ ID NO: 44, SEQ ID NO: 45, SEQ ID NO: 46, SEQ ID NO: 47, SEQ ID NO: 48,
SEQ ID NO: 49, SEQ ID NO: 50, SEQ ID NO: 51, SEQ ID NO: 52, SEQ ID NO: 53,
SEQ ID NO: 54, SEQ ID NO: 55, SEQ ID NO: 56, SEQ ID NO: 57, SEQ ID NO: 58,
SEQ ID NO: 59, SEQ ID NO: 60, SEQ ID NO: 61, SEQ ID NO: 62, SEQ ID NO: 63,
SEQ ID NO: 64, SEQ ID NO: 65, SEQ ID NO: 66, SEQ ID NO: 67, SEQ ID NO: 68,
10 SEQ ID NO: 69, SEQ ID NO: 70, SEQ ID NO: 71, SEQ ID NO: 72, SEQ ID NO: 73,
SEQ ID NO: 74, SEQ ID NO: 75, SEQ ID NO: 76, SEQ ID NO: 77, or SEQ ID NO: 78.

161. The method of claim 1, wherein the capture oligonucleotide comprises the
sequence set forth in SEQ ID NO: 17, SEQ ID NO: 18, SEQ ID NO: 19, SEQ ID NO: 20,
15 SEQ ID NO: 21, SEQ ID NO: 22, SEQ ID NO: 23, SEQ ID NO: 24, SEQ ID NO: 24,
SEQ ID NO: 26, SEQ ID NO: 27, SEQ ID NO: 28, SEQ ID NO: 29, SEQ ID NO: 30,
SEQ ID NO: 31, SEQ ID NO: 32, SEQ ID NO: 33, SEQ ID NO: 34, SEQ ID NO: 35,
SEQ ID NO: 36, SEQ ID NO: 37, SEQ ID NO: 38, SEQ ID NO: 39, SEQ ID NO: 40,
SEQ ID NO: 41, SEQ ID NO: 42, SEQ ID NO: 43, SEQ ID NO: 44, SEQ ID NO: 45,
20 SEQ ID NO: 46, SEQ ID NO: 47, SEQ ID NO: 48, SEQ ID NO: 49, SEQ ID NO: 50,
SEQ ID NO: 51, SEQ ID NO: 52, SEQ ID NO: 53, SEQ ID NO: 54, SEQ ID NO: 55,
SEQ ID NO: 56, SEQ ID NO: 57, SEQ ID NO: 58, SEQ ID NO: 59, SEQ ID NO: 60,
SEQ ID NO: 61, SEQ ID NO: 62, SEQ ID NO: 63, SEQ ID NO: 64, SEQ ID NO: 65,
SEQ ID NO: 66, SEQ ID NO: 67, SEQ ID NO: 68, SEQ ID NO: 69, SEQ ID NO: 70,

SEQ ID NO: 71, SEQ ID NO: 72, SEQ ID NO: 73, SEQ ID NO: 74, SEQ ID NO: 75,
SEQ ID NO: 76, SEQ ID NO: 77, or SEQ ID NO: 78.

162. The method of claim 38, wherein at least one of the capture
5 oligonucleotides comprise the sequence set forth in SEQ ID NO: 17, SEQ ID NO: 18,
SEQ ID NO: 19, SEQ ID NO: 20, SEQ ID NO: 21, SEQ ID NO: 22, SEQ ID NO: 23,
SEQ ID NO: 24, SEQ ID NO: 24, SEQ ID NO: 26, SEQ ID NO: 27, SEQ ID NO: 28,
SEQ ID NO: 29, SEQ ID NO: 30, SEQ ID NO: 31, SEQ ID NO: 32, SEQ ID NO: 33,
SEQ ID NO: 34, SEQ ID NO: 35, SEQ ID NO: 36, SEQ ID NO: 37, SEQ ID NO: 38,
10 SEQ ID NO: 39, SEQ ID NO: 40, SEQ ID NO: 41, SEQ ID NO: 42, SEQ ID NO: 43,
SEQ ID NO: 44, SEQ ID NO: 45, SEQ ID NO: 46, SEQ ID NO: 47, SEQ ID NO: 48,
SEQ ID NO: 49, SEQ ID NO: 50, SEQ ID NO: 51, SEQ ID NO: 52, SEQ ID NO: 53,
SEQ ID NO: 54, SEQ ID NO: 55, SEQ ID NO: 56, SEQ ID NO: 57, SEQ ID NO: 58,
SEQ ID NO: 59, SEQ ID NO: 60, SEQ ID NO: 61, SEQ ID NO: 62, SEQ ID NO: 63,
15 SEQ ID NO: 64, SEQ ID NO: 65, SEQ ID NO: 66, SEQ ID NO: 67, SEQ ID NO: 68,
SEQ ID NO: 69, SEQ ID NO: 70, SEQ ID NO: 71, SEQ ID NO: 72, SEQ ID NO: 73,
SEQ ID NO: 74, SEQ ID NO: 75, SEQ ID NO: 76, SEQ ID NO: 77, or SEQ ID NO: 78.

163. The method of claim 38, wherein at least one of the target nucleic acid
20 sequences is a portion of a gene of a *Staphylococcus* bacterium and at least one of the
target nucleic acid sequences is a portion of the *mecA* gene.

164. The method of claim 38, wherein the method is used to distinguish
between two or more species of a common genus.

25

165. The method of claim 164, wherein the species differ by two or more non-consecutive nucleotides.

166. The method of claim 164, wherein the species differ by two or more
5 consecutive nucleotides.

167. The method of claim 164, wherein the species differ by at least one nucleotide.

10